4. Fine mapping of a pollen semi-sterility gene in rice (*Oryza sativa* L.)

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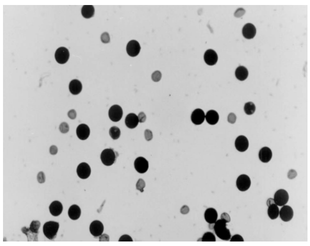
So far, it is not clear for the molecular mechanism of the partial sterility in the *indica*/*japonica* hybrids. Many difficulties existed in studying the genetic mechanism for the partial sterility caused by *indica*/*japonica* crosses, which may be solved by using stable semi-sterility rice mutant.

A spontaneous semi-sterile mutant named as W207-2 was found from a *japonica* rice variety Nipponbare, in which pollen fertility was about 50 percent and spikelet fertility about 40 percent (Fig. 1). The pollen and spikelet fertility of W207-2, and other parents as well as reciprocal F_1 hybrids was shown in Table 1. The results suggested that the semi-sterility was controlled by recessive nucleus genes with no cytoplasmic effect.

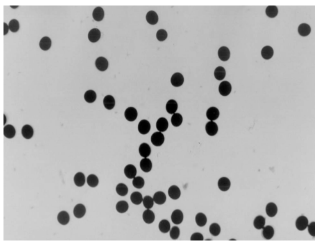
Ten spikelets of W207-2 were successively pollinated with the pollen of W207-2 and Nipponbare for 4 days, respectively. The spikelet fertility pollinated with the pollen of W207-2 was 50.45%, and still semi-sterility, however, the spikelet fertility pollinated with the pollen of Nipponbare was 85.55%. It implied that the female fertility of W207-2 was normal. By analyzing the 291 plants in W207-2/Nipponbare F_2 population, positively significant linear correlation (γ=0.9119) was obtained between the pollen and spikelet fertilities at 1% level. These results suggested that the spikelet semi-sterility was mainly caused by pollen semi-sterility. At the same time, we found that the anther of W207-2 was thinner and almost indehiscent, while that of Nipponbare was plumper and dehiscent. It suggested that the anther indehiscence was also affected the spikelet semi-sterility of W207-2.

Frequency distribution of pollen fertility in the W207-2/Nipponbare F_2 population was continuous and bimodal. According to the apparent low valley (70%), the W207-2/Nipponbare F_2 population could be divided into two groups, the fertile plants group including 221 individuals and the semi-sterile plants group including 70 individuals. χ^2 tests showed that fertile plants and semi-sterile plants fitted to 3:1 ratio (χ^2_c=0.0928). These results suggested that the pollen semi-sterility of W207-2 might be controlled by a single recessive nucleus gene.

To determine the locus for the semi-sterile gene, the fertile and semi-sterile bulks were made by mixing equal amounts of DNA from 20 fertile and semi-sterile plants in the W207-2/Dular F_2 population, respectively. Then RM506, RM152, RM6356 and RM6863 on chromosome 8 were detected polymorphisms between the fertile and semi-sterile bulks as well as the two parents, and likely to be linked to *pss1* loci. Thus, the linkage map of chromosome 8 was constructed by assaying 182 random plants of W207-2/Dular F_2 population. Only one major QTL controlling pollen semi-sterility (*pss1*) with a LOD score 48.3 and a PVE (phenotypic variation explained) of 70.5% was identified at the region between RM6356 and a new developed markers RS41 on chromosome 8, and confirmed by analysis of another F_2 population from W207-2/Nipponbare. In previous study, Sobrizal et al. (2001) mapped a pollen semi-sterile gene, *S27(t)*, between G2132 and L128 on chromosome 8. Yu et al. (2005) detected a pollen and spikelet semi-sterile gene, *sst(1)*, between RM152 and RM6863, which corresponded well with *pss1*. Furthermore, *pss1* was finely mapped between SSR makers RS41 and RS43, and co-segregated with RS413 using 449 recessive individuals. The map distance between *pss1* and RS41 as well as RS43 was both 0.3cM. Both RS41 and RS43 markers were located on the same PAC clone P0470F10, with the physical distance of about 125kb (Fig. 2). Thus this study lays a foundation for map-based cloning the *pss1* gene. The finding of the pollen semi-sterile gene will be useful for understanding the molecular genetic mechanism of rice semi-sterility.



A



B

Fig. 1. Pollen grain fertility. A: W207-2. B: Nipponbare.

Table 1. Pollen and spikelet fertilities of the parents and reciprocal F_1

Varieties and F_1s	Pollen fertility（%）	Spikelet fertility（%）
Nipponbare	97.0±0.5	94.4±1.1
W207-2	51.0±1.2	40.1±1.1
Dular	98.6±0.7	91.7±1.7
W207-2/Nipponbare	91.1±1.7	87.5±1.4
Nipponbare/W207-2	93.2±1.9	88.1±1.6
W207-2/Dular	90.9±1.6	87.1±1.3
Dular/W207-2	94.6±1.9	89.5±1.2

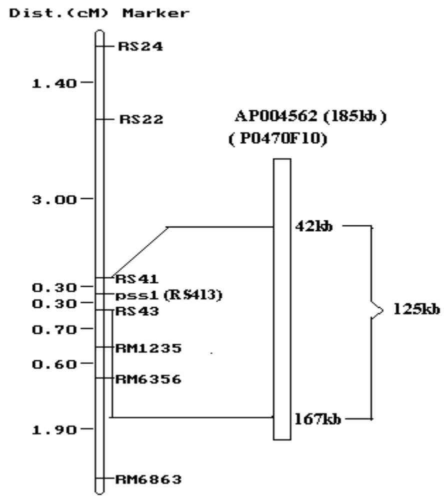


Fig. 2. Linkage map of the region included *pss1* on chromosome 8.

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